Effect of Agitation of EDTA with 808-Nanometer Diode Laser on Removal of Smear Layer

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Abstract

Introduction: Near-infrared diode lasers can be used for several applications, which range from disinfection to smear layer removal in endodontics. This study evaluated the efficacy of agitation of 15% EDTA with an 808-nm diode laser on removal of the smear layer. **Methods:** Sixty extracted human maxillary central incisor teeth were instrumented up to ProTaper F4 (Dentsply Maillefer, Ballagues, Switzerland) and then randomly divided into 6 groups (n = 10 for each group) according to the different final irrigating protocols as follows: 5% sodium hypochlorite for 120 seconds performed with the NaviTip (Dentsply Maillefer, Ballaigues, Switzerland) (control group); 15% EDTA for 120 seconds performed with the NaviTip; and agitation of 15% EDTA with an 808-nm diode laser for 10, 20, 30, and 40 seconds. Specimens were observed under a scanning electron microscope, and open dentinal tubules were counted using Adobe Photoshop software (Adobe Systems, San Jose, CA). The data were analyzed with 1-way analysis of variance and Tukey post hoc tests (P = .05). **Results:** The number of open dentinal tubules was higher in the middle thirds than in the apical thirds. The differences between the apical and middle thirds were statistically significant (P < .05). Statistically significant differences were also found between the control group and the other groups in both the middle and apical thirds of the root canals (P < .05). **Conclusions:** The results indicated that agitation of 15% EDTA with an 808-nm diode laser for 20 seconds was effective in removing the smear layer in the apical thirds of root canals. *(J Endod 2013;39:1589–1592)*

Key Words

Agitation, diode laser, EDTA, laser, smear layer

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The chemomechanical preparation of root canal dentine with hand or rotary instruments creates debris and a smear layer (1). The smear layer is composed of organic and inorganic components, such as vital or necrotic pulp tissue, microorganisms, saliva, blood cells, and dentinal debris (2). It has an amorphous and irregular appearance and consists of 2 separate layers. Because it can become infected and can harbor bacteria and bacterial products (3, 4), the smear layer should be removed. However, irrigating solutions can be inefficient in removing the smear layer from the apical thirds of root canals (5, 6). Researchers have proposed improved irrigation agitation methods to address this issue (7). Recently, agitation of irrigating solutions with laser devices has become popular (8–10). In a study on the penetration of sealers in curved root canals with and without agitation using a laser, the agitation of sodium hypochlorite (NaOCl) resulted in a significantly higher amount of sealer penetration than in the nonagitated group. However, the study found no significant differences in sealer penetration for EDTA irrigation between the nonagitated group and the laser-agitated group (10).

The near-infrared diode laser has some advantages; it has a thin and flexible fiber, which allows access into narrow and curved root canals, and it provides increased disinfection of the deep radicular dentin (11, 12). To date, no study has investigated the effect of agitation with an 808-nm diode laser on the removal of the smear layer. The aim of this study was to evaluate the efficacy of agitation of 15% EDTA with an 808-nm diode laser at different agitation time intervals. The null hypothesis was that the laser-agitated irrigation with EDTA would not increase the efficacy of 15% EDTA in removing the smear layer.

Materials and Methods

Laser System

An 808-nm wavelength gallium-aluminum-arsenide diode laser (Doctor Smile, Lambda Scientifica Srl, Vicenza, Italy) system equipped with a 20-W power source was used. The laser delivery system used in the study was a fiber-optic cable with a 300-μm size at 2 W in the pulsed mode (10 ms on/10 ms off). The actual power of the parallel fiber-optic tip was 285.71 W/cm^2.

Specimen Preparation

The protocol of this study was approved by the Research Ethics Committee of Izmir Katip Celebi University, Izmir, Turkey. The study included 60 single-rooted, noncarious maxillary human central incisor teeth that had been freshly extracted because of periodontal or prosthodontic reasons from patients of both sexes ranging in age from 45–55 years. After extraction, the intact apical root tips and the presence of a single root canal were verified with buccopalatinal and mesiodistal radiographs. Caution was paid to make sure all root canals had similar initial apical file sizes (#15) and root canal anatomy (straight). A periodontal scaler mechanically removed soft tissues and calculus from the surfaces of the roots. The specimens were disinfected in a 0.5% solution of chloramine T (Merck, Darmstadt, Germany) for 48 hours and then stored in 4°C distilled water until use (13). The specimens were decoronated with a diamond disc (Diamond Disc Superflex 910S/220; North Bel, Paderno Dugnano, Italy) under water coolant to obtain a standardized root length of 15 mm.

The pulp tissues were extirpated using a barbed broach (VDW, Munich, Germany), and a size #10 stainless steel K-file (Dentsply, Maillefer, Ballaigues,

Switzerland) was moved down in the canal until the file was just visible. The working lengths were set by deducting 1 mm from these lengths. The apexes of the specimens were closed with boxing wax.

The root canals were shaped with ProTaper rotary instruments (Dentsply Maillefer, Ballagues, Switzerland). The root canals of all the specimens were apically prepared up to size #40 (F4). They were irrigated with 2 mL 5% NaOCl (ImidentMedEndosolve-HP, Konya, Turkey) between instrument changes. All the irrigation procedures were performed with 31-G side port irrigator tips (NaviTip; Ultradent, South Jordan, UT). During the irrigating procedures, the irrigator tips were placed 1 mm from the working length, and they were then moved backwards and forwards. After the preparations of root canals, the specimens were divided randomly into the following 6 groups of 10 teeth each:

1. *Control group:* The specimens in this group were irrigated with 5 mL 5% NaOCl for 120 seconds followed by 10 mL distilled water. The total volume of the irrigants was 15 mL.
2. *EDTA group:* The specimens in this group were irrigated for 120 seconds with 5 mL 15% EDTA (Wizard; Rehber Kimya San, Istanbul, Turkey) as a final flush, and the specimens were then irrigated with 5 mL 5% NaOCl for 120 seconds followed by a final rinse with 5 mL distilled water. The total volume of the irrigants was 15 mL.
3. *10 seconds of agitation with the laser:* In this group, 1 mL 15% EDTA solution was placed in the canals of the specimens and then agitated at 2 W using the pulsed mode (10 Ton ms/Toff 10 ms) of the diode laser for 10 seconds. The specimens were then irrigated for 110 seconds with 4 mL 15% EDTA as a final flush and irrigated with 5 mL 5% NaOCl for 120 seconds followed by a final rinse with 5 mL distilled water.
4. *20 seconds of agitation with the laser:* In this group, 2 mL EDTA was agitated at 2 W using the pulsed mode of the diode laser for 20 seconds as in group 3. The specimens were irrigated for 100 seconds with 3 mL 15% EDTA as a final flush and then irrigated with 5 mL 5% NaOCl for 120 seconds followed by a final rinse with 5 mL distilled water.
5. *30 seconds of agitation with the laser:* In this group, 3 mL EDTA was agitated at 2 W for 30 seconds using the pulsed mode of the diode laser. The specimens were irrigated for 90 seconds with 2 mL 15% EDTA as a final flush and then irrigated with 5 mL 5% NaOCl for 120 seconds followed by a final rinse with 5 mL distilled water.
6. *40 seconds of agitation with the laser:* In this group, 4 mL EDTA was agitated at 2 W in the pulsed mode of the diode laser for 40 seconds. The specimens were irrigated for 80 seconds with 1 mL 15% EDTA as a final flush and then irrigated with 5 mL 5% NaOCl for 120 seconds followed by a final rinse with 5 mL distilled water.

The total irrigant volume was standardized to 15 mL in all groups. The exposure time to the 15% EDTA was also standardized at 120 seconds. All the laser applications were performed parallel to the root canal. The tip of the laser was kept 2 mm from the working length and was withdrawn gently from the apical region to the coronal region with a helical movement. The roots were then dried with absorbent paper points. The boxing waxes were removed, and the specimens were stored in 2.5% glutaraldehyde solution buffered with phosphate for 24 hours to obtain fixation. After 24 hours, the specimens were rinsed under tap water, and 2 parallel grooves were made on the buccal and palatal surfaces using a diamond disk followed by splinting and immersion in liquid nitrogen. The specimens were dehydrated using a series of graded ethanol solutions (70%, 80%, 90%, and 100%) for 24 hours. Finally, they were placed in an oven at 37°C for 48 hours. One half of each specimen was selected and prepared for scanning electron microscopic evaluation. The specimens were coated with a gold-palladium layer (Polaron SC7610; Quorum Technologies Ltd, East Sussex, UK) and analyzed with a digital scanning electron microscope (EVO LS10; Zeiss, Oberkochen, Germany) at 10-mm working length. Two photomicrographs were taken from the middle and apical thirds of the root canals at a magnification of 3000×. The open dentinal tubules were counted on each photomicrograph using Adobe Photoshop software (Adobe Systems, San Jose, CA) (Fig. 1). Statistical analyses were performed using SPSS software (SPSS Inc, Chicago, IL). The Kolmogorov-Smirnov statistical test for normality revealed a normal

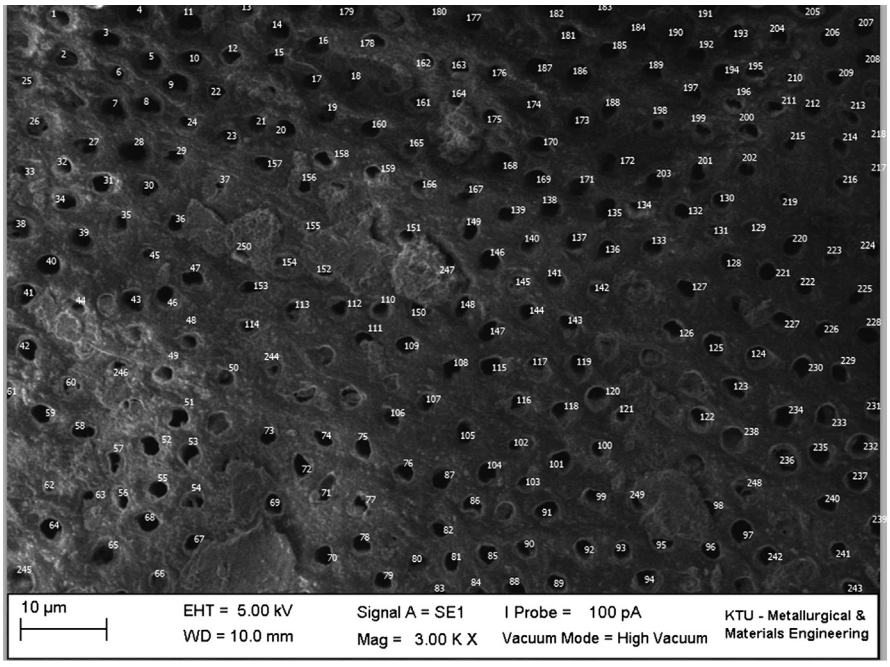

Figure 1. Representative photomicrograph shows the counting of the open dentinal tubules.

data distribution. Statistical analysis was performed using 1-way analysis of variance and Tukey post hoc tests ($P = .05$).

Results

The number of open dentinal tubules in 120 photomicrographs was counted using Adobe Photoshop software. The means and standard deviations of the open dentinal tubules in the middle and apical thirds were 126.5 ± 91.09 and 51.87 ± 58.70, respectively. According to the statistical analysis, the middle thirds contained a higher number of open dentinal tubules than the apical thirds of the root canals ($P < .001$).

In the middle third of the root canals, the means and standard deviations of the open dentinal tubules of the control; EDTA; and agitation with the laser for 10, 20, 30, and 40 seconds groups were 5.7 ± 3.59, 149.3 ± 69.49, 154.8 ± 46.3, 166.3 ± 77.02, 154.6 ± 115.63, and 128.30 ± 88.23, respectively. Fewer open dentinal tubules were observed in the control group, and the highest number was observed in the 20 seconds of agitation with the laser group. According to the statistical analysis, both the EDTA and the agitation with the laser (10, 20, 30, and 40 seconds) resulted in more open dentinal tubules compared with the control group ($P < .001$) (Fig. 2).

In the apical third of the root canals, the means and standard deviations of the open dentinal tubules of the control; EDTA; and agitation with the laser for 10, 20, 30, and 40 seconds groups were 3.2 ± 1.47, 47 ± 47.08, 56.6 ± 54.72, 91.9 ± 44.92, 66.2 ± 82.25, and 46.34 ± 62.22, respectively. Fewer open dentinal tubules were observed in the control group, and the highest number was observed in the 20 seconds of agitation with the laser group. According to the statistical analysis, the EDTA and 10, 30, and 40 seconds of agitation with the laser groups resulted in a similar number of open dentinal tubules compared with the control group ($P > .05$). However, the number of open dentinal tubules in the 20 seconds of agitation with the laser group was higher ($P = .008$) compared with the control group.

Discussion

Effective chemomechanical preparation of the apical region is especially important for successful root canal treatment. Syringe irrigation is a standard procedure for root canal irrigation, but this technique is not efficient in the apical third of the root canal. It is difficult to completely remove the residual smear layer, particularly in the apical third of the root, because the smaller size of the apical third compared with the other thirds impedes the circulation and action of the irrigating solutions (5, 6). In addition, increased tubular sclerosis in the apical third means there are fewer dentinal tubules (14–18). Acoustic and hydrodynamic properties of irrigants have been studied to improve the effectiveness of irrigating solutions in the apical region (19); agitation with a laser has been used in endodontic therapy to reduce the number of bacteria and to modify the surface of the root canal (20). Therefore, the present study investigated whether agitation with a diode laser improves the efficacy of EDTA, especially in the middle and apical thirds.

The absorption of a root canal irrigant depends on the type of irrigating solution and the wavelength of the laser. In a recent study by Moon et al (10), activation with a 1320-nm Nd:YAG laser with NaOCl or EDTA was found to be much better than NaOCl for sealer penetration into dentinal tubules. It has been shown that the absorption of EDTA at 1320 nm is higher than at the 810-nm wavelength (21). The mechanism for the laser activation of irrigating solutions originates from the absorption of laser energy, the formation of vapor bubbles, the collapse of the bubbles, acoustic streaming, and finally cavitation. In the present study, an 808 nm diode laser was used with 15% EDTA. Interestingly, in the middle and apical thirds of root canals, the highest number of open dentinal tubules was observed in the group with 20 seconds of agitation using the laser. However, the water absorption of the 808-nm diode laser is poor. Thus, no comparison can be made between the results of the studies in which other laser types were used.

Clinically, the root is enclosed by the bone socket, and the canal behaves as a closed-end channel. This situation results in gas entrainment at the end of this region, producing a vapor lock effect during the

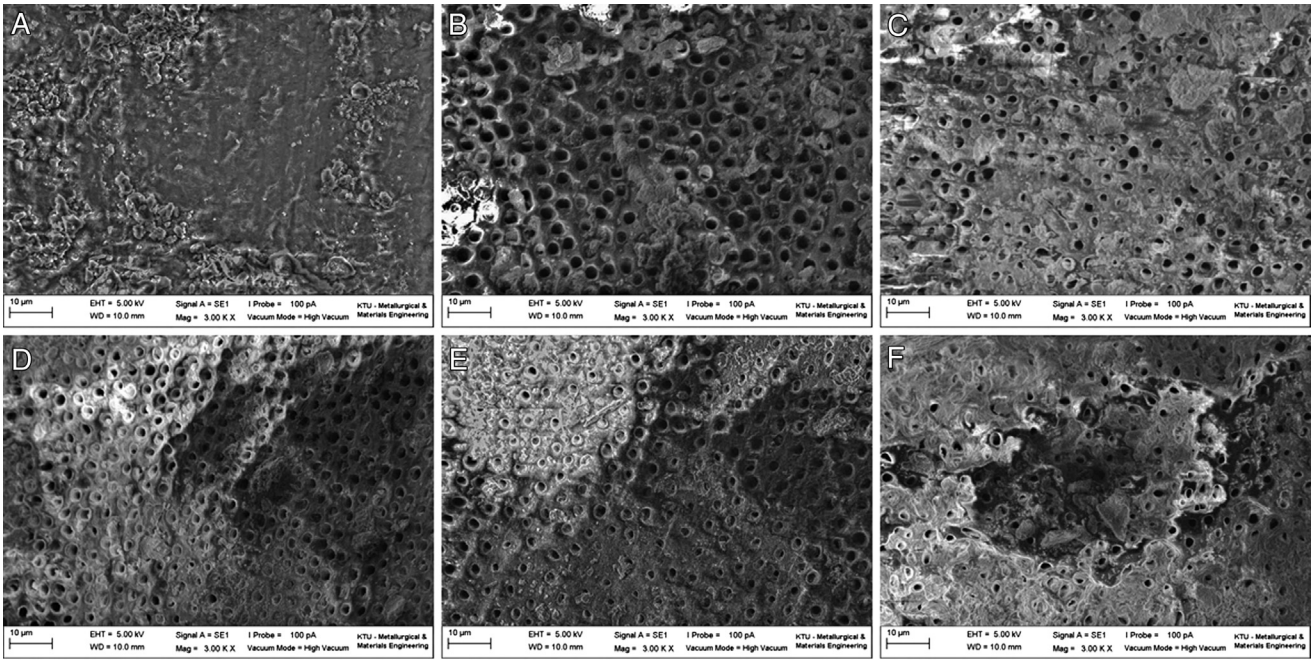

Figure 2. Representative photomicrographs of groups: (*A*) group 1: no EDTA irrigation, (*B*) group 2: 15% EDTA, (*C*) group 3: 10 seconds of agitation with the laser, (*D*) group 4: 20 seconds of agitation with the laser, (*E*) group 5: 30 seconds of agitation with the laser, and (*F*) group 6: 40 seconds of agitation with the laser.

irrigation procedure. This could be why there was more smear layer removal from the apical thirds in the group that received agitation using the diode laser for 20 seconds than in the control group. Future studies should be conducted to confirm the results of the present study.

One limitation of this study is that the temperature of the EDTA before and after lasing was not measured. The protocols of the study were standardized, such as the exposure time of the EDTA, the volume of the EDTA, and the concentration of the EDTA. However, the agitated EDTA solution volume was not standardized (ie, the aim was to agitate the fresh solution at every turn). Thus, 1 mL EDTA in each 10-second laser application was agitated. This was another limitation of the study. A further limitation of this study was the small sample size, which resulted in high standard deviations.

To inhibit possible irrigant extrusion, Matsuoka et al (22) proposed that 200- or 320-mm fibers should be kept 2–3 mm away from the anatomic apex to hinder eradication of the apical constriction. In the present study, the laser fiber tip was applied 2 mm away from the anatomic apex to avoid extrusion.

A previous study reported that the amount of smear layer removed by EDTA in 30 seconds was poor (23). This finding is in accordance with another study, which applied EDTA for 1 minute (24). In the present study, EDTA was applied in all groups for 120 seconds.

Scanning electron microscopy has been used to determine the effectiveness of agitated irrigants to eliminate the smear layer. This technique makes it possible to examine the morphologic features of the surfaces of prepared root canals. Various systems have been used to score the amount of smear layer remaining after root canal preparation. Score systems were not used in the present study. Instead, the number of open dentinal tubules was counted by trained observers with software. The software provides a viable and objective alternative to conventional methods (25).

Conclusion

The null hypothesis was rejected. Within the limitations of this study, it can be concluded that 20 seconds of agitation of EDTA with an 808-nm diode laser improved the efficacy of EDTA in removing the smear layer.

Acknowledgment

The authors deny any conflicts of interest related to this study.

References

1. Violich DR, Chandler NP. The smear layer in endodontics–a review. Int Endod J 2010;43:2–15.
2. Czonstkowsky M, Wilson EG, Holstein FA. The smear layer in endodontics. Dent Clin North Am 1990;34:13–25.
3. Pashley DH. Smear layer: physiological considerations. Oper Dent Suppl 1984;3:13–29.
4. McComb D, Smith DC. A preliminary scanning electron microscopic study of root canals after endodontic procedures. J Endod 1975;1:238–42.
5. Torabinejad M, Cho Y, Khademi AA, et al. The effect of various concentrations of sodium hypochlorite on the ability of MTAD to remove the smear layer. J Endod 2003;29:233–9.
6. Mancini M, Armellin E, Casaglia A, et al. A comparative study of smear layer removal and erosion in apical intraradicular dentine with three irrigating solutions: a scanning electron microscopy evaluation. J Endod 2009;35:900–3.
7. Guerisoli DM, Marchesan MA, Walmsley AD, et al. Evaluation of smear layer removal by EDTAC and sodium hypochlorite with ultrasonic agitation. Int Endod J 2002;35:418–21.
8. De Moor RJ, Blanken J, Meire M, et al. Laser induced explosive vapor and cavitation resulting in effective irrigation of the root canal. Part 2: evaluation of the efficacy. Lasers Surg Med 2009;41:520–3.
9. de Groot SD, Verhaagen B, Versluis M, et al. Laser-activated irrigation within root canals: cleaning efficacy and flow visualization. Int Endod J 2009;42:1077–83.
10. Moon YM, Kim HC, Bae KS, et al. Effect of laser-activated irrigation of 1320-nanometer Nd:YAG laser on sealer penetration in curved root canals. J Endod 2012;38:531–5.
11. Wang X, Sun Y, Kimura Y, et al. Effects of diode laser irradiation on smear layer removal from root canal walls and apical leakage after obturation. Photomed Laser Surg 2005;23:575–81.
12. de Souza EB, Cai S, Simionato MR, et al. High-power diode laser in the disinfection in depth of the root canal dentin. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2008;106:68–72.
13. De-Deus G, Brandao MC, Leal F, et al. Lack of correlation between sealer penetration into dentinal tubules and sealability in nonbonded root fillings. Int Endod J 2012;45:642–51.
14. Druttman AC, Stock CJ. An *in vitro* comparison of ultrasonic and conventional methods of irrigant replacement. Int Endod J 1989;22:174–8.
15. Ram Z. Effectiveness of root canal irrigation. Oral Surg Oral Med Oral Pathol 1977;44:306–12.
16. Teixeira CS, Felippe MC, Felippe WT. The effect of application time of EDTA and NaOCl on intracanal smear layer removal: an SEM analysis. Int Endod J 2005;38:285–90.
17. Paque F, Luder HU, Sener B, et al. Tubular sclerosis rather than the smear layer impedes dye penetration into the dentine of endodontically instrumented root canals. Int Endod J 2006;39:18–25.
18. Abou-Rass M, Patonai FJ. The effects of decreasing surface tension on the flow of irrigating solutions in narrow root canals. Oral Surg Oral Med Oral Pathol 1982;53:524–6.
19. Weller RN, Brady JM, Bernier WE. Efficacy of ultrasonic cleaning. J Endod 1980;6:740–3.
20. George R, Meyers IA, Walsh LJ. Laser activation of endodontic irrigants with improved conical laser fiber tips for removing smear layer in the apical third of the root canal. J Endod 2008;34:1524–7.
21. Meire MA, Poelman D, De Moor RJ. Optical properties of root canal irrigants in the 300-3,000-nm wavelength region. Lasers Med Sci 2013;27:1307–14.
22. Matsuoka E, Jayawardena JA, Matsumoto K. Morphological study of the Er, Cr:YSGG laser for root canal preparation in mandibular incisors with curved root canals. Photomed Laser Surg 2005;23:480–4.
23. Prado M, Gusman H, Gomes BP, et al. Scanning electron microscopic investigation of the effectiveness of phosphoric acid in smear layer removal when compared with EDTA and citric acid. J Endod 2011;37:255–8.
24. Serper A, Calt S. The demineralizing effects of EDTA at different concentrations and pH. J Endod 2002;28:501–2.
25. George R, Rutley EB, Walsh LJ. Evaluation of smear layer: a comparison of automated image analysis versus expert observers. J Endod 2008;34:999–1002.